THE METHOD OF CLEAVAGE IN THE SPORANGIA OF CERTAIN FUNGI

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[WITH PLATES 15 AND 16 AND TEXT-FIGURES A–F]

The division of the multinucleated sporangia into spores and the delimitation of the sporangium from the sporangiophore by a dome-shaped cross partition present many features of special cytological interest. Erroneous views on both processes are still current in the literature and as to the formation of the columella certain textbooks, both old and recent, are still entirely out of harmony with the facts. The literature dealing with the common molds, *Mucor* and *Rhizopus,* constitutes some of the earliest contributions to mycological research.

Corda (11), the "father of microscopic mycology," gave us the first description and illustration of the method of the columella and spore formation in *Ascophora mucedo,* now known as *Rhizopus nigricans.* He writes: "Nun rundet sich die kolbige Verdickung allmälig, und gleichzeitig erfüllt die gelbe Masse ihren Hohlenraum völlig . . . aus dem unteren Theile mit dem Stiele verbunden beginnt allmälig die polsterartige Erhebung der Columella, und gleichzeitig beginnt die obere Masse undeutliche und noch isolirte Zellen zu bilden welche sich vermehren und endlich in enger Verbindung die ganze Sporangie erfüllen." His figure 78 (5–9) depicts the columella as at first slightly arched and gradually pushing up into the sporangium, while the spores are represented, at their very inception, as polyhedral masses. These figures of Corda are doubtless responsible for the persistent, erroneous accounts of the columella formation still to be found in certain textbooks, though Corda's statements were soon contradicted by Fresenius.

Fresenius (20), also studying *Rhizopus nigricans,* states he was unable to observe the phenomena described and figured by Corda. He writes: "Was über Bau und Entwickelung der Sporen bei Corda gesagt und abgebildet ist scheint mir völlig aus der Luft gegriffen zu sein." Further, also as early as 1872, Brefeld (7),

studying *Mucor mucedo,* made the following statement: "Die Scheidewand ist nicht etwa ursprünglich horizontal und erhällt ihre gewölbte Form durch Dehnung unter dem einfluss des Druckes der Flüssigkeitssaüle im Fruchtträger, wie mehrfach angegeben wird. sie hat in der ersten Anlage die gewölbte Gestalt, die nachträglich nur unwesentlich modificirt wird."

Mucor mucedo and *Rhizopus* have been used as types not only in advanced and elementary textbooks of botany, but in many so-called textbooks of biology. Here especially the desire to elaborate in detail on the life processes beyond what is in the literature has led to many false statements, which should certainly be corrected in the interest of sound teaching. Illustrations of such incorrect statements are found in the following texts:

Bessey (4) figures and describes the development of the sporangium of *Mucor mucedo* as follows: ". . . the vertical hyphae which are filled with protoplasm become enlarged at the top and in each a transverse partition forms (*A, a,* fig. 159), the portion above the partition (*b,* fig. 159) becomes larger, and at the same time the transverse partition arches up (*B, a,* fig. 159), finally appearing like an extension of the hypha, and is then called the columella (*C, a,* fig. 159)." Reynolds Green (21), referring to *Mucor mucedo,* states: "A septum is formed close to the apex of the hypha, cutting off a small head, which grows and becomes globular. The lower cell grows also and projects into the swollen portion, forming a columella." Parker (40), referring to *Mucor,* states: ". . . The sporangium continues to grow, and as it does so the septum becomes more and more convex upwards, finally taking the form of a short club-shaped projection, the columella, extending into the interior of the sporangium." Atkinson (1) writes: ". . . at the same time that this end cell is enlarging the cross wall is arching up into the interior. This forms the columella." Coulter, Barnes and Cowles (13), referring to the Mucorales, write: "After the terminal sporangium cell is cut off, the separating wall bulges into the sporangium cavity, forming the so-called columella." Bigelow (5) incorporates in his text Parker's faulty figure of the method of the columella formation. Nathansohn (38) states: ". . . durch eine Querwand schnürt sich an deren Spitze eine

Zelle ab, schwillt zur Kugel an, in deren Hohlraum sich die Querwand meist mehr oder weniger einstülpt und die sog. Columella bildet." In so recent a textbook as Densmore's General Botany (17) we find: "This sporangial cell now expands with great rapidity and with its expansion the wall separating it from its hyphal stalk grows in surface area and assumes a convex form, protruding into the growing sporangium until it comes to occupy fully one half or two thirds of the sporangial cavity, when it is called the columella." Densmore also figures (fig. 141) the columella as at first a plane wall, which is later arched up into the sporangium.

The method of spore formation in sporangia was studied, with interesting results, as early as 1859, following the discovery of cell formation by division, as worked out by Von Mohl and others, and has been prosecuted up to the present day. While the pioneers were influenced and sometimes misled by theories relative to cell formation in general, the fact remains that as early as 1859 Pringsheim (44) observed and figured progressive cleavage from the surface inward, essentially as we know the process today, in the sporangia of *Lagenidium entophytum* (Pringsheim), Zopf (*Pythium entophytum* Pringsheim). He writes as follows: "Erst vor der Oeffnung des Sporangium beginnt nun in dem ausgetretenen, zur Kugel zusammen gebalten Inhalt, eine an der Peripherie beginnende und nach dem Centrum vorschreitende Sonderung durch welche die Protoplasmakugel schliesslich in eine grössere Anzahl von Schwarmsporen zerfällt [Pl. 8, fig. 1*b*]."

General conclusions relative to spore formation in sporangia found in some recent papers are quite at variance with observations which seem well established by earlier students. One must infer that some of the recent writers must have overlooked Rothert's (46) paper entitled, "Die Entwicklung der Sporangien bei den Saprolegnieen."

The general history of this literature has been treated by Swingle (50) and recently by Moreau (37) and Harper (25). I shall refer only to points bearing on matters that seem still unsettled, especially the question as to the occurrence of so-called simultaneous cell-division.

Van Tieghem (55–56–57), in a series of papers dealing practically with the entire group of Zygomycetes, undoubtedly laid the foundation for the later conception of simultaneous division. In reference to *Sporodinia,* he writes as follows: "Le protoplasma sporigene se separe d'abord en deux substances tres differente. La premiere toujours granuleuse, se condense en petites portions polyedriques qui deviendront bientot autant de spores."

Twenty years later Leger (34), in his very fully illustrated thesis, dealing with fourteen species of Zygomycetes, quotes Van Tieghem's reference to the manner of spore formation in *Sporodinia* and adds: "En somme, ce passage montre d'une facon tres exacte le developpement des spores dans ses traits principaux." The discovery of cell-plates in the division of the cells of the higher plants undoubtedly influenced the conclusions of many students of spore formation.

Strasburger (48–49) for *Saprolegnia* and *Mucor mucedo,* Büsgen (8) for the Saprolegniales, *Phytophthora, Cystopus* and *Mucor mucedo,* Ward (59) for *Phytophthora infestans,* and Maurizio for *Olpidiopsis* state that the cell-division in these forms is by cell-plates.

Fischer (19), studying spore formation in the sporangia of *Woronina,* describes the process as follows: "der Zerfall des Sporangiumplasma in eine der grosse desselben entsprechende Anzahl anfangs polyedrischen Portionen, die zukünftigen Schwärme." Van Tieghem, as noted, refers to a condensation into polyhedral portions. Fischer observes a breaking up into polyhedral masses. It is interesting to note that the same author (Fischer (19)) regarded the spore plasm of *Olpidiopsis* and *Rozella* as suddenly forming rounded spores. In the former case he writes: "Mit einem male zerfällt das gesammte Inhalt in scharf umschriebene rundliche Theilchen. . . ." In the latter case he states: "Plötzlich zerfällt nun in einem gegebenen momente das Protoplasma in eine menge rund umschriebenen Portionen die zukünftigen Zoosporen." Pringsheim (43–45) regards the spore formation as occurring "unmittelbar" (directly) in *Achlya prolifera, Olpidiopsis, Rozella,* and *Woronina.*

Strasburger (48) was perhaps the first to use the term, simul-

taneous, as describing spore formation: "In den Zoosporangien der Saprolegnien wird, wie aus zahlreichen angaben bekannt, eine grosse Anzahl Schwarmsporen simultan aus dem gesammten Protoplasmatischen Inhalte des sporangium gebildet."

Dangeard (14), studying spore formation in *Synchytrium taraxaci,* calls it simultaneous fragmentation. Popta (42), who was concerned with the question of periplasm and spore formation in the so-called Hemiasci, refers to spore production in *Protomyces bellides* as "Simultan und sehr rasch." Barrett (3), investigating some species of *Olpidiopsis,* states that segmentation of the sporangial contents is apparently simultaneous throughout.

Cornu (12), studying the Chytridiales, parasitic on Saprolegniales, refers to spore formation in *Olpidiopsis* as follows: "Presque sans transition, le contenu s'organise en petites masses spheriques, futures zoospores." He claims that a similar phenomenon occurs in the sporangia of *Rozella* and *Woronina.* Thus in the abovementioned genera the spore plasm is said to organize, with practically no transitional stages, into spherical masses.

Büsgen, who, as already mentioned, made a comparative study of a number of Saprolegniales, Peronosporales, and Mucorales, combines the conceptions of cell-plates and division, not always simultaneous. He says: "Unter auftreten von Zellplatten theilt sich der gesammte Inhalt des Sporangiums—nicht immer simultan—in etwa gleich grosse, meist nicht regelmässig begrenzte Portionen. . . ."

Rothert (46), in a quite thorough piece of work on *Saprolegnia,* figured clefts from the central vacuole proceeding outward. Humphrey (29), studying the Saprolegniales of America, merely states his agreement with Rothert as to cleavage and spore formation.

Thaxter (51) finds spore formation simultaneous in *Syncephalastrum,* but occurring by progressive constriction in *Syncephalis pycnosperma, S. nodosa, S. Wynneae, S. cordata,* and in a *Syncephalis* closely allied to the latter.

Kusano (32) and Griggs (23) hold that spore formation in *Synchytrium puerariae* and *Rhodochytrium,* respectively, may be either simultaneous or progressive. Davis (15), studying spore formation in the sporangia of *Saprolegnia,* Wager (58) in *Poly-*

phagus Euglenae, and Butler (9) in *Pseudolpidium aphanomycis* record spore formation as proceeding from the center to the periphery by cleavage, but do not refer to it as progressive. Likewise, Hartog (27), investigating *Pseudospora Lindstedii,* a monadine parasitic on *Saprolegnia,* figures cleavage by vacuoles extending to the periphery of the protoplasmic mass, but does not refer to it as progressive. Davis (16) figures progressive cleavage in the sporangia of the alga, *Derbesia,* by means of furrows starting from the periphery and proceeding inward. Loewenthal (35) has studied spore formation in *Olpidium dicksonii* and *Zygorhizidium willei* and Griggs (22) has studied *Monochytrium,* but both authors leave the question unsettled whether the cleavage is progressive or simultaneous.

In 1899, Harper (24), studying cell-division in sporangia and asci, pointed out that in the sporangia of *Synchytrium decipiens* the cleavage is accomplished by furrows, which form on the surface of the initial cell, and by growing deeper in a more or less radial fashion divide the protoplasmic mass, successively, into smaller portions. Harper also investigated the spore and columella formation in *Pilobolus crystallinus* and *Sporodinia grandis.* He finds, as Brefeld had stated, that the columella is not first a plane wall, which is eventually pushed up into the sporangium, but that it is from the first dome-shaped, a layer of vacuoles appears near the inner boundary of the dense spore plasma, which subsequently flatten and fuse and thus delimit the spore plasma from the columella plasma. In the case of *Pilobolus* the columella formation is aided by cleavage furrows cutting in at the base of the sporangium.

In *Pilobolus,* as in *Synchytrium decipiens,* the cleavage is progressive and is initiated by the formation of surface furrows which deepen and finally cut the plasma into protospores. In *Synchytrium decipiens* the uninucleated protospores become multinucleated and enlarge to form the spores which in germination again become sporangia. In *Pilobolus* the progressive cleavage leads to the formation of one or few nucleated protospores. These protospores become multinucleated, increase in size, and divide until finally oblong, binucleate sporangiospores are produced. In *Sporodinia,* Harper finds an abbreviated process of spore formation in that the

progressive cleavage, by surface furrows and clefts, divides the spore plasm into multinucleated, polygonal blocks of very variable size which round up at once and become the definitive spores.

Swingle (50) finds cleavage in the sporangia of *Rhizopus nigricans* much like that in *Sporodinia,* except that the spores are more uniform in size and have thicker walls. In *Phycomyces* the spore plasm is divided by vacuoles, which become angular and fuse to form irregular clefts. Spore formation is aided by furrows which cut into the spore plasm from the columella cleft. Swingle agrees with Harper as to the method of columella formation.

Timberlake (52) describes spore formation in the sporangia of the alga, *Hydrodictyon,* as a progressive cleavage by means of furrows. Percival (41) and Rytz (47), discussing spore formation in *Synchytrium endobioticum* and *Synchytrium succissae,* respectively, both agree that spore formation is brought about by progressive cleavage.

In 1913, Moreau (37) described the spore formation in a number of Zygomycetes. His study may be summarized as follows:

In *Circinella conica* spore formation proceeds by means of vacuoles, which separate fragments of protoplasm having the form of amoebae. The protoplasm contracts around each nucleus, rarely around two nuclei, forming protospores which he compares to those described by Harper for *Pilobolus.* The nuclei then divide and lead to the formation of multinucleated spores. Moreau states that for *Phycomyces nitens* and *Rhizopus nigricans* his observations agree in general with those of Swingle, but on page 32 he refers to the protoplasmic segments as being "amiboide" and connected by trabeculae. In *Mucor spinescens,* Moreau finds that a confluence of vacuoles leads to the formation of elongated protoplasmic threads. The threads become nodose, each nodosity containing one or two nuclei and finally forming a spore. Moreau states that a similar thread stage may be observed in the spore formation in *Absidia glauca* and *Absidia septata.* In *Syncephalastrum cinereum* and *Syncephalastrum racemosum* the spores are said to be formed by a condensation of protoplasm into spherical or elliptical masses, each enclosing one or more spores; generally there is but one nucleus in each spore. Moreau's description of

spore formation, in the above-mentioned zygomycetes, is extremely fragmentary and certain of his figures suggest that his material was poorly fixed.

METHODS

The *Saprolegnias* were grown on small flies of the genus *Drosophila*. They were frequently parasitized by *Olpidiopsis* and *Rozella*. Cultures on the flies were then transferred to slightly cooled agar plates. A drop of water was then placed on each fly so that the sporangiferous filaments might float out into their normal position. Slightly cooled agar was then gently dropped over each fly. The cultures were then exposed out-of-doors to quickly congeal the agar. The halo of filaments was still easily discernible. Blocks of agar containing the entire host were now cut out and transferred to weak Flemming and Merkel fixatives. The washing, dehydrating, and imbedding was done as usual. The sections were cut 5 μ thick, stained with the Flemming triple combination, cleared very quickly in clove oil, and mounted in Canada balsam.

Cleavage in *Saprolegnia* was also studied in hanging-drop cultures, and *Olpidiopsis* was also studied in the same manner. The Zygomycetes were cultured upon sterilized bread in jelly glasses. In order to retain the loose, open structure of the bread, which facilitates the growth of the mycelia, only a small amount of water was poured into each jelly glass before sterilization. When the sporangia assumed a snow-white appearance, under the hand-lens, wefts of the fungus were cut out with sharp-pointed scissors and immediately transferred to the fixatives. The conglomerated mass of hyphae was then gently pushed down into the fixative and the vial was shaken to dislodge the air-bubbles. The material was fixed for 24–48 hours in Merkel's solution, or for one hour in one part of weak Flemming and two parts of water, and then transferred to Merkel's fixative. By these means blackening of the fungus was prevented and bleaching with hydrogen peroxide was unnecessary.

The fixative was now poured off and the vial was carefully filled with water and tilted into a dish of water. The fungus was repeatedly floated into a vial and transferred into fresh water. The material, thus washed for two to two and one half hours, was

then dehydrated, beginning with 15% alcohol, and imbedded in paraffin. The sections were cut 5 μ thick, stained by Flemming's triple method, cleared quickly in clove oil, and mounted as usual.

OLPIDIOPSIS

Pringsheim (45) gave us the first account of spore formation in the sporangia of *Olpidiopsis,* a parasite, which he mistook for the antheridia of *Saprolegnia.* He speaks of the spores as "Samenkörper" and says that they are formed directly (unmittelbar), and that similar phenomena may be observed in the structures which we now recognize as the sporangia of *Rozella* and *Woronina.*

In 1872, Cornu (12) published a paper in which he supported A. Braun (6) in reference to the parasitic nature of *Olpidiopsis.* He noted the appearance of large centrally disposed vacuoles, their disappearance, and the formation of a foamy protoplasm. Both Cornu (12) and Fisher (19) agree that the spores are formed directly.

Maurizio (36) states that cell-plates are formed in spore formation in *Olpidiopsis major.* As already mentioned, Loewenthal is not clear as to whether spore formation in *Olpidium dicksonii* and *Zygorhizidium willei* is simultaneous or by progressive cleavage.

In Butler's (9) account of spore formation in *Pseudolpidium aphanomycis* he states that the spore "Anlage" originate as a result of "heapings of protoplasm," which are few in number as compared with the number of zoöspores produced. Butler compares this stage with that Harper describes in *Synchytrium,* where the early stages of cleavage give rise to multinucleated masses of protoplasm. Cleavage fissures then extend from the vacuole to the sporangial wall, the vacuolar and protoplasmic membranes then rupture, and the "Anlage" swell and fuse. The sporangium is now filled with a homogeneous mass. Butler states: "Five or ten minutes later final fashioning of the zoospores is complete and movement commences in the sporangium." Butler records cleavage furrows extending from the vacuole to the sporangium wall, but he does not figure them. His figure 6*C,* plate 9, which he interprets as a "condensation of protoplasm into heaped masses,"

appears to be a stage prior to the enlargement and fusion of the vacuoles into a large central vacuole.

Barrett (3) studied both living and stained material of a number of *Olpidiopsis* species. He reports that he could not detect any signs of the protoplasmic heapings described by Butler within the sporangium. If one studies the figures in plate 24, one is led to believe that Barrett did not find the crucial stages of spore formation. This leads him to the erroneous conclusion that "fragmentation of the protoplasm is simultaneous."

Kusano (33) investigated the life history and cytology of *Olpidium viciae*. He was unable to find evidence of progressive division, but states "that a clear space appeared in the cytoplasm all at once between each two nuclei, and that the protoplasm was cut up into as many polygonal parts as there were nuclei." It seems obvious that this statement refers to the stage following what Strasburger and Büsgen regarded as a stage of coalescence of the spore origins which is not real, but only apparent.

The stages in the life history of *Olpidiopsis saprolegniae,* prior to the formation of spores, have been discussed and figured by a number of authors. From a study of the living material, in hanging-drop cultures, I am able to confirm the existence of large centrally disposed vacuoles in young sporangia, the increment in size, and subsequent coalescence of these vacuoles. The process of spore formation, as I have observed it, is entirely at variance with that described by the above-mentioned authors. The phenomena I observed were very similar to processes of spore formation in the sporangia of *Saprolegnia* and *Achlya* as I have found them and not as described by Strasburger, Büsgen, and Hartog. I was able to note the following changes by observing living specimens in hanging-drop cultures. The history of one sporange is as follows:

At 7:40 P.M. the sporange had a large central vacuole, and in the median plane a blunt cleavage furrow could be seen extending toward the periphery (text fig. A, 4). Three minutes later the vacuole became irregular and one sharp and two blunt cleavage furrows were visible (text fig. A, 5). Four minutes later the vacuole had increased in size so that the protoplasm formed a rather thin peripheral layer. Sharp cleavage furrows were now

evident (text fig. A, 6, 7), and after the elapse of one minute they cut through the latter and the large vacuole disappeared. Cleavage is now complete and here and there the outlines of the spores can be made out (text fig. A, 8). After two minutes the protoplasm became very granular and the hazy outlines of polygonal spore masses were recognizable. I can only interpret this polygonal stage as due to rapid growth of the spore initials, which thus press against one another and become polyhedral. One minute after the appearance of the compact, polygonal spore initials a contraction occurred, the inter-sporal substance appearing as hyaline lines.

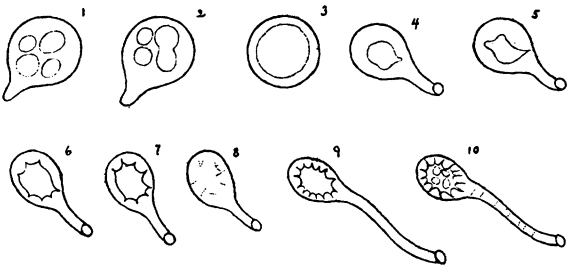

Fig. A. *Olpidiopsis saprolegniae:* 1–2, median view of sporangia showing several rounded vacuoles; 3, sporangium showing coalescence of the vacuoles; 4–8, different series than 1–3 in which the wall layer is thinner; 4–5, sporangia showing vacuoles of various shapes; 6–7, sporangia showing early cleavage stages; 8, sporangium showing apparent homogeneous stage following the rupture of the plasma membrane; 9-10, another individual, sporangia show radial furrows; 10, cleavage has occurred in the exit tube. 1–2 and 4–10 show exit tubes.

One minute later the spores underwent a further contraction; they rounded up and almost immediately began to move to and fro. Within two minutes the zoöspores escaped through the exit tube. Thus within eighteen minutes of the first formation of a cleavage furrow the spores formed and escaped. If one considers the rapidity of these changes, one can readily infer why the cleavage furrows, extending outward from a central vacuole, are so seldom seen in fixed sections. I can not agree, therefore, with Barrett that the spore formation occurs simultaneously. I would interpret his figure 39 as a contraction stage following the so-called homogeneous state, of Strasburger and Büsgen, in which the spore initials are so closely pressed together that their boundaries are

almost obliterated. Nor can I agree with Butler, who describes the formation of "protoplasmic heapings" and the delimitation of multinucleated, protoplasmic masses, which are later cut up by cleavage furrows, to form the spore origins. In no case did I observe any fusion of spore initials and the resulting production of a homogeneous state such as Butler describes for *Pseudolpidium aphanomycis* and as was held by Strasburger and Büsgen to occur in *Saprolegnia.*

We can summarize, roughly, the following stages in the spore formation of *Olpidiopsis saprolegniae* Cornu:

1. Protoplasm with many small vacuoles.
2. The formation of large vacuoles more or less centrally disposed and the concomitant production of an exit-tube.
3. The coalescence of large vacuoles into a large central vacuole.
4. Progressive cleavage by furrows cutting outward from the central vacuole. (First contraction phase.)
5. Cleavage of plasma membrane, shrinkage of the sporangium and disappearance of the central vacuole.
6. Swelling of the spore initials to the polygonal closely pressed areas commonly observed.
7. Second contraction phase—appearance of hyaline spaces between spore initials (often erroneously interpreted as cell plates).
8. Further contraction leading to the rounding up of the spore masses and their swarming movements.

The above conclusions were reached after carefully studying the cleavage phenomena in dozens of sporangia in hanging-drop cultures.

SAPROLEGNIA AND ACHLYA

Rothert (46) recognized three types of sporangia in the Saprolegniaceae: "gefüllte Sporangien," those completely filled with protoplasm; "inhaltsarme," those having a thin parietal layer of protoplasm; and "normale," sporangia with a thick parietal layer, the predominant form. Rothert figured furrows cutting through the protoplasm from the central vacuole outward and notes that these furrows appear practically simultaneously throughout the whole length of the sporangium. Rothert's observations on spore formation in *Saprolegnia* and *Achlya* are of great importance for understanding the cleavage phenomena in the other sporangia. Harper has reviewed and confirmed Rothert's observations in sev-

eral points, but recent students have in a number of cases failed to take account of the evidence he has presented as to the contraction and expansion phases accompanying cleavage. I have studied *Achlya* and *Saprolegnia* in both living, sectioned, and stained material, and my observations confirm those of Rothert.

The process of cleavage is similar to that I have already described for *Olpidiopsis saprolegniae*. The cleavage is progressive, the furrows appear first on the inner surface of the parietal protoplasmic layer and give the latter an undulated appearance. Gradually these clefts become sharper and reach the plasma membrane. In optical view these protoplasmic masses resemble the old-fashioned sugar-loaves. Viewed from the surface the protoplasmic masses are roughly polygonal. The spore initials are generally described as being connected by fine protoplasmic strands. I am inclined to interpret these strands as gelatinous exudates of the spore initials. Rothert described the development of spores in very slender sporangia as a heaping of protoplasm on the protoplasmic membrane. It is a question whether there is much, if any, increase in the radial diameter of the protoplasmic layer on the median axis of the spore initials. A better interpretation of the spore formation in these sporangia and in the oögonia is to regard it as a process of cleavage, the furrows being at first broad and shallow instead of sharp and deep.

The spore initials now contract and the clefts become prominent. The protoplasmic masses now become densely granular, are highly refractive, and assume more definite outlines. This stage is quickly followed by a splitting of the protoplasmic membrane which is drawn in by the isolated spore initials as they round up. Division of the protoplasmic content is now complete, each definitive spore is uninucleated and is homologous with the uninucleated protospores Harper described in *Synchytrium decipiens*.

As first noted by Rothert and confirmed by Harper, the splitting of the elastically stretched plasma membrane is attended with a marked shrinkage of the sporangium wall accompanied by the expulsion of part of the cell-sap through the sporangial wall. The basal septum, which has heretofore been concave, is now pushed up by turgor into the sporangium and assumes a convex configura-

tion. Rothert estimated the shrinkage at 13 per cent. Butler (9) finds that at a corresponding stage the diameter of the sporangium of *Pythium intermedium* decreased by about one tenth. The same author also noted a contraction of the sporangium of *Pseudolpidium aphanomycis* immediately after the disappearance of the large central vacuole. Harper called attention to the fact that in *Synchytrium decipiens* the cleavage was accompanied by pronounced shrinkage. He found that when cleavage was complete the total volume of the segments had been reduced to such an extent as not to occupy more than approximately one third of the volume of the primordial cell. Kusano also reports a shrinkage of the segments in the sporangia of *Synchytrium puerariae* during the cleavage process.

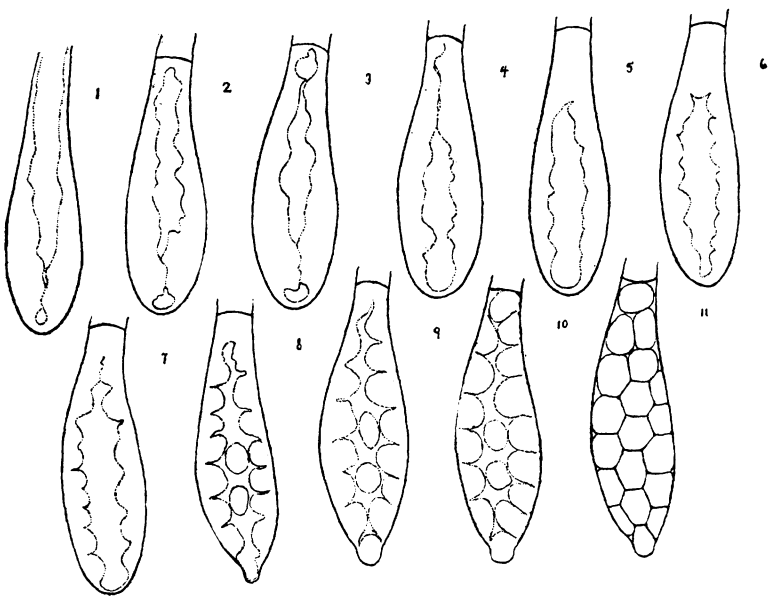

Fig. B. *Saprolegnia torulosa:* 1, tip of hypha which will become a sporangium; 2–7, show varying appearances of the central vacuole; 7, first appearance of cleavage, furrows irregular and not corresponding to the definitive furrows; 8–9, spore masses outlined by rather shallow furrows, the two oval outlines in these figures, and also in figures 10, represent the end views of spore initials projecting up from below; 10, clefts have become deeper; 11, spores have become polygonal through mutual pressure; this stage soon follows the rupture of the plasma membrane and represents an expansion phase of the spores; the basal wall is now convex toward the sporangium showing that its plasma membrane is ruptured.

Text figure B, 1–11, represents the lengths and breadths of a sporange at successive stages in spore formation. The micrometer measurements are as follows:

9:12 P.M.	86 μ long—26 μ wide
9:27 P.M.	88 μ long—26 μ wide
9:37 P.M.	90 μ long—25 μ wide
9:56 P.M.	92 μ long—25 μ wide
10:05 P.M.	93 μ long—25 μ wide
10:10 P.M.	94 μ long—28 μ wide
10:17 P.M.	90 μ long—27 μ wide
10:22 P.M.	92 μ long—26 μ wide
10:30 P.M.	86 μ long—22 μ wide
10:34 P.M.	Spores moved to and fro.

At 10:10 P.M. the sporange had reached its maximum size; the basal wall was concave, due to the turgor within the sporange. At 10:17 P.M. the clefts apparently cut through the plasm membrane, the spore initials rounded up, and the sporange decreased four microns in length and one micron in diameter. The basal wall was now flattened. This stage of contraction was followed by the expansion stage. The spore initials became tightly pressed together, the protoplasm assumed a homogeneous appearance, and the spore outlines were only visible as very faint lines. This is the stage that deceived Strasburger (48), for he writes as follows: "Wiederholt sind mir Fälle vorgekommen in welchen nach dem die Sporenanlage es schon bis zur Bildung der Körnergrenzen ja selbst Hautschichtgrenzen gebracht hatte, plötzlich die ganze Entwickelung rückgängig wurde, alle Trenungsandeutungen schwanden und das Sporangium alsbald wieder von gleichmässig kammerigen Protoplasm gefüllt erschien. Dann nach kurzer Zeit, wurde die Entwickelung, und zwar nun auffallend schnell wieder aufgenommen. Eine solche zweite fiel mir, im Verhältniss zu der Ersten stets durch die grosse Regelmässigkeit der Theilstücke auf."

The great expansion following the delimitation of the spore initials, the temporary loss of the granular character of the spore protoplasm, the obscuring of the cell boundaries through close contact, and the subsequent contraction which reveals the polygonal spore masses have given rise to much confusion. Butler (9), in 1907, speaking of spore formation in *Pythium proliferum*, writes: "From this I have been led to suppose that even at this stage the

spore origins are definitely formed, and that, though fused into a mass in which individual spores can not be made out, yet each nucleus has obtained a hold on a certain mass of cytoplasm. . . ."

The sudden appearance of the polygonal spore masses at the beginning of the second contraction phase has given rise to such theories as the simultaneous cleavage of the sporangial protoplasm into polygonal masses and the cutting out of the spores by cell-plates. I have already summarized these views and I need not repeat them here.

The polygonal spores of *Saprolegnia* undergo a further contraction and subsequently round up. The turgor in the sporangium is decreased to such an extent that the basal wall now becomes convex inward. At this stage the sporangium decreased still more in length. Thus during the period of greatest turgidity the sporangium measured 94 microns in length. When the spores were fully matured the sporangium had contracted eight microns in length and six microns in width.

The observations of Rothert (46) relative to the escape of the cell-sap and the concomitant shrinking of the sporangium during spore formation in *Saprolegnia,* Harper's evidence of similar phenomena in *Synchytrium decipiens,* Kusano's observation of shrinkage in *Synchytrium pucrariae,* Swingle's account of progressive cleavage in *Rhizopus nigricans* and *Phycomyces nitens,* Harper's studies of spore formation in the sporangia of *Sporodinia grandis* and *Pilobolus,* Butler's observations relative to shrinkage in the sporangia of *Pythium intermedium* and *Pseudolpidium aphanomycis,* Harper's and Dodge's observations of the extrusion of water into vacuoles during the early stages of the formation of sporangia in *Trichia,* as well as my own observations, lead me to corroborate the contention of Harper that the exudation of water is a factor in the process of segmentation of the protoplasm. Harper compares the furrowing of the spore plasm with the cracking of a drying, colloidal mass. The fact that vacuoles or furrows never cut out protoplasmic segments devoid of nuclei is proof that the latter are the centers which control the water loss and thus the cleavage process. This may be explained by assuming that the nucleic acids manifest an attraction or affinity for water greater

than that displayed by the cytoplasm; hence, as Harper has suggested, the loss of water may be least in the vicinity of the nuclei.

SPORODINIA GRANDIS

Spore formation in the sporangia of *Sporodinia grandis* was regarded by both Van Tieghem (55) and Leger (34) as a condensation of the spore plasm into polyhedric masses, which later round up. Harper (24) has figured a number of stages in spore and columella formation. Swingle (50), a few years later, studied the same fungus and reports that his results are entirely in accord with those of Harper.

As *Sporodinia* represents an extreme type as to the speed of spore formation, I have studied the process further in the light of the conceptions of contraction and expansion first developed by Rothert from his studies on the sporangia of *Saprolegnia.* I find the dome formed by series of large vacuoles, which flatten, fuse, end to end, and separate the spore plasm from the columella plasm as described by Harper. I have, however, a number of slides (Pl. 15, figs. 1, 6) which show an interesting variation of the process in that the vacuoles are completely fused on one side of the sporangium, while on the other side they are either somewhat globose or flattened. Swingle's fig. 8, plate 2, shows that in *Rhizopus nigricans* the columella formation may be more advanced on one side of the sporangium. A few times I observed surface furrows cutting in at the base of the sporangium, to meet the flattened vacuoles, which cut out the columella (Pl. 15, fig. 6). Harper has described a similar phenomenon in *Pilobolus* and Swingle in *Rhizopus nigricans.*

In *Sporodinia* spore formation may begin before the columella cleft is complete. Swingle notes that this often occurs in *Rhizopus nigricans* (see his fig. 8, plate 2). This shows, it seems to me, that the columella formation and cleavage are two parts of one general contraction phase. The cleavage is progressive and may begin by the formation of furrows at the surface or at the columella cleft (Pl. 15, figs. 1, 2). It is to be noted that cleavage is more advanced in that region where the fusion of vacuoles has produced the columella cleft (Pl. 15, fig. 6). The furrows cut

inwardly and as the spore plasm is giving off water the clefts widen. The furrows from the surface appear to cut into the spore plasm in a centripetal fashion. They meet and fuse with those furrows which started from the columella cleft and cleavage is thus completed; the protoplasm has been cut up into a mass of irregular blocks which are variable in size and are multinucleate. These spore initials represent, as compared with swarm spores of *Saprolegnia* and the protospores of *Synchytrium decipiens, Pilobolus crystallinus, Circinella conica,* etc., a premature completion of spore formation. They correspond to the multinucleate masses preceding the protospores. As in *Rhizopus nigricans,* the spores of *Sporodinia grandis* are multinucleate at their inception. When cleavage is complete the spore initials present a dense granular appearance. The protoplasmic mass is also somewhat shrunken. Soon, however, the spore initials take up water and grow, the protoplasm becomes less granular and takes on a lighter stain. The spores become so tightly pressed together that their protoplasmic membranes assume polyhedral outlines, which are so thin that they are traced with difficulty under the oil-immersion objective (Pl. 15, fig. 4). This stage is homologous with the so-called homogeneous stage, which Strasburger and Büsgen described for *Saprolegnia.* This period of growth is followed by a second contraction. The spores now develop a thin wall, contract slightly, and round up.

I have illustrated the chief stages of the development of the columella and spores in *Sporodinia grandis* in a series of text figures (text fig. C, 1–5).

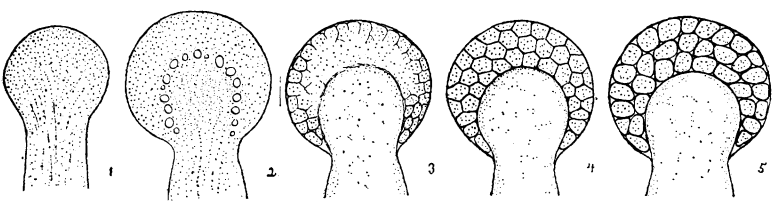

Fig. C. Diagrams showing the method of the columella and spore formation in *Sporodinia grandis,* the nuclei appearing as mere dots. 1, a young sporangium. 2, showing the dome-shaped layer of vacuoles outlining the columella. 3, showing early stage of progressive cleavage. 4, showing the polyhedral stage. 5, mature spores.

MUCOR RACEMOSUS

Apparently no one since Leger (34) has studied spore formation in *Mucor racemosus*. Leger claims for it, as for all sporangia, that the spores are cut out simultaneously as polyhedric blocks.

Moreau (37) has studied *Mucor spinescens* and he describes vacuolization of the protoplasm resulting in long strands, which become nodular and then break up into uninucleate or several nucleated spores. He gives no further account of the process.

I have studied *Mucor racemosus* relative to the process of spore formation. I find that spore formation is initiated by furrows, which start as the periphery of the spore plasm and cut out multinucleated blocks of irregular size (Pl. 16, fig. 16). Further furrowing cuts up these blocks into irregular protoplasmic masses containing a few nuclei. These protoplasmic masses then grow and become polyhedral. This expansion stage is followed by a second contraction, the spores round up and develop a cell-wall. The mature spores may contain seven to eight nuclei. I have not observed a protospore stage. The process of spore formation, as in *Sporodinia grandis,* is abbreviated, but the spores have thicker walls and are viable for a longer period. They are also more uniform in size than those of *Sporodinia grandis.*

CIRCINELLA MINOR

Moreau (37), studying spore formation in *Circinella conica,* states that the center of the sporangium is at times occupied by a large vacuole. The formation of spores is accomplished by irregular vacuoles, which cut up the spore plasm into amoeba-like bodies bound together by protoplasmic strands. These strands become thinner and break, the protoplasm then contracts about each nucleus, rarely around two. Moreau compares these protoplasmic bodies to the protospores Harper described in *Pilobolus crystallinus.* The nucleus then divides and each protoplasmic mass becomes multinucleated, the spores become polygonal and press against one another. At maturity they become globular and smooth.

I have studied spore formation in *Circinella minor,* but my observations do not agree entirely with those of Moreau. Cleavage,

as I have observed it, is similar to that Harper described in *Pilobolus.* Furrows appear at the surface of the spore plasm and cut inwardly to meet the clefts produced in the interior of the spore plasm by vacuoles, which become angular (Pl. 16, fig. 8). The spore plasm is thus cut up into irregular protoplasmic blocks containing a variable number of very small nuclei (Pl. 16, fig. 8). Moreau does not describe or figure cleavage furrows in *Circinella conica.* The irregular blocks are further divided by cleavage into more or less oblong to sausage-shaped protoplasmic masses containing four to five nuclei (Pl. 16, fig. 9). As in *Pilobolus,* these blocks are transversely divided into roughly polygonal, one- to two-nucleated protoplasmic masses. I agree with Moreau in calling these protoplasmic segments the protospores. During the cleavage process the protoplasmic mass undergoes shrinkage without question, but I did not observe such a loose and open structure of the dividing spore plasm as Moreau figures, and I am inclined to believe that his figure 28, plate 3, represents poor fixation and considerable shrinkage. The protospores are, for a time, connected by delicate, gelatinous strands, which are probably an exudate of the protoplasm (Pl. 16, fig. 10). The nuclei now divide and each protoplasmic mass (protospore) swells and grows. The young spores now become polyhedral and are closely pressed together (Pl. 16, fig. 13). This expansion period is followed by a contraction; the multinucleated spores round up and form a cell-wall (Pl. 16, fig. 14).

The process of spore formation in *Circinella minor* may be summarized as follows:

1. Differentiation of spore and columella plasm.
2. Formation of irregular multinucleate blocks of protoplasm by surface furrows and angular vacuoles.
3. Further division by cleavage producing oblong protoplasmic masses containing four to five nuclei (2 and 3 are contraction phases).
4. Division of oblong to sausage-shaped blocks into one- to two-nucleated protospores.
5. Protospores grow and become multinucleated (expansion phase).
6. Spores round up (second contraction phase).
7. Further contraction and formation of cell-walls.

Harper has pointed out that in *Sporodinia grandis* there is an abbreviation of the process of spore formation as compared with

Pilobolus crystallinus and *Synchytrium decipiens.* It is evident that the process of spore formation in *Circinella minor,* like *Rhizopus nigricans,* occupies an intermediate position in such a series. In *Circinella minor* the formation of protospores is followed by nuclear division and growth. But with the formation of the protospores cell-division is complete. In *Pilobolus crystallinus* the protospore grows and becomes multinucleated, but this multinucleated cell divides by constriction. The final cell-divisions produce the oblong, binucleate spores. In *Sporodinia grandis* the process of spore formation is so abbreviated that the initial cleavage cuts out comparatively large multinucleated segments which ultimately round up and become the definitive spores.

I have illustrated the chief stages of the development of the columella and spores in *Mucor mucedo* in a series of text figures (text fig. D, 1–5).

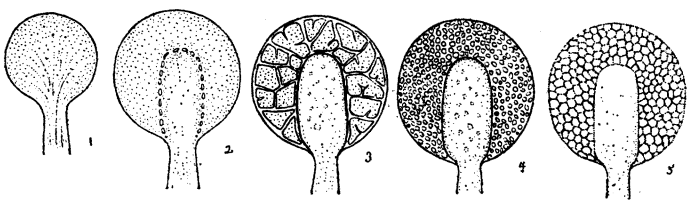

Fig. D. Diagrams showing the method of the columella and spore formation in *Mucor mucedo.* 1, a young sporangium. 2, showing the dome-shaped layer of vacuoles outlining the columella. 3, showing the spore plasm cut up into protoplasmic blocks by progressive cleavage. 4, spore-initials in the contraction stage. 5, showing the polyhedral or expansion stage.

MUCOR MUCEDO

Strasburger (49) has given us a fragmentary account of spore formation in *Mucor mucedo.* He considered the protoplasmic mass cut up by cell-plates in a manner similar to that in *Saprolegnia.* Two years later Büsgen (8), studying the same species, came to the conclusion that the spore plasm is cut up into large protoplasmic blocks by cell-plates, and that by subsequent subdivision protoplasmic masses are formed which have the size of the definitive spores. The sporangium then becomes homogeneous and a *second division* produces the definitive spores. Leger (34) studied spore formation in the sporangia of *Mucor mucedo* and

agrees with Van Tieghem that the spore plasm is divided at once into polyhedric granular spores separated by a non-granular substance. Moreau,[1] studying spore formation in *Mucor mucedo,* came to the conclusion that the spore plasm divides into irregular multinucleated fragments which subsequently become the spores.

I have studied the method of the columella and spore formation in *M. mucedo.* I find that the columella does not originate as a plane wall, which is subsequently arched up into the sporangium, as is so often depicted in textbooks on botany, but that as in the Zygomycetes studied by Harper (24) and Swingle (50) the columella is from the first dome-shaped as I show in Pl. 16, fig. 19; a dome-shaped series of vacuoles appear, these flatten, fuse end to end, and thus delimit the spore plasm from the columella plasm.

The spore plasm is first cut up into comparatively large protoplasmic blocks. During this stage considerable contraction occurs for the blocks are not in close apposition (Pl. 16, fig. 20). These blocks are now subdivided by cleavage into roughly polyhedral spore initials. This subdivision is attended by still further contraction, followed by an expansion stage in which the spore initials become polygonal, as figured by Leger ((34), plate 8, fig. 35; my figure, plate 16, fig. 21). These spore initials eventually contract and form the ovate definitive spores. I have not been able to determine with certainty the number of nuclei in the ripe spores.

RHIZOPUS NIGRICANS AND PILOBOLUS CRYSTALLINUS

The process of spore formation in *Sporodinia grandis* is much abbreviated, the spore plasm being cut up only into relatively large multinucleate blocks (text fig. C, 1–5), which quickly round up to form the definitive spores. In *Rhizopus nigricans* (text fig. E, 1–5) the spore plasm is cut up, progressively, into numerous much smaller multinucleate, angular to ovate spores, but never reaches the uninucleate stage. The relative extent of the cleavage is well illustrated by comparing the size of the spores of *Rhizopus nigricans* with that of those of *Sporodinia grandis.* In *Sporodinia grandis* the spores measure, on an average, 20–30 x 17–24 μ, in *Rhizopus nigricans* 9–12 x 7.5–8 μ. In *Pilobolus crystallinus* (text fig. F, 1–6) the process of spore formation is still further

[1] Bull. Soc. Mycol. Fr. 31: 71–72. 1915.

protracted. The spore plasm is cut up, by progressive cleavage, into uninucleate protospores. An embryonic stage now intervenes, the protospores grow and become multinucleate. By a series of divisions binucleate definitive spores are produced.

For the sake of comparison I have also included diagrams of *Rhizopus nigricans* and *Pilobolus crystallinus,* showing stages of the development of the columella and spores.

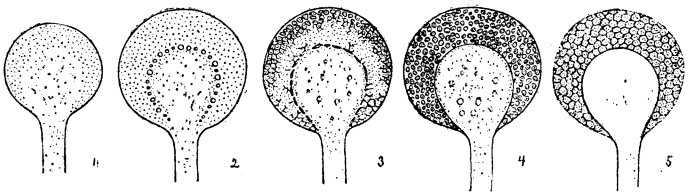

Fig. E. Diagrams showing the method of the columella and spore formation in *Rhizopus nigricans.* 1, a young sporangium. 2, showing the dome-shaped layer of vacuoles outlining the columella. 3, showing early stage of cleavage. 4, showing the contraction stage. 5, expansion or polyhedral stage.

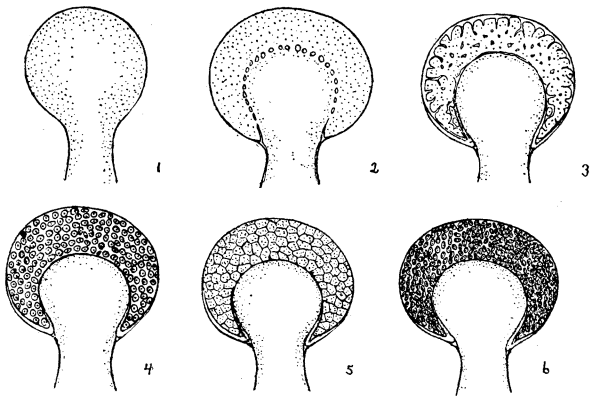

Fig. F. Diagrams showing the method of the columella and spore formation in *Pilobolus crystallinus.* 1, a young sporangium showing dense spore plasm. 2, showing the dome-shaped series of vacuoles and a circular furrow which cut out the columella. 4, stage of uninucleate protospores. 5, polyhedral stage. 6, ripe spore stage.

DISCUSSION

Although a number of recent papers by Kusano (32), Barrett (3), and Griggs (23) report the occurrence of simultaneous cleavage in the sporangia of certain algae and fungi, the evidence to

prove the existence of this method of spore formation is inadequate. On the other hand, there is an accumulation of evidence which confirms the contention that cell-division in the sporangia of algae and fungi is essentially a process of furrowing either from the periphery of the sporangia or from the vacuoles in the interior of the spore plasm.

As far as I am aware Rothert, studying spore formation in the sporangia of *Achlya* and *Saprolegnia,* was the first to note the contraction and expansion phases during the cleavage process. His observations are, therefore, of paramount importance for understanding the mechanics of the cleavage phenomena in other sporangia.

A complete parallelism with the phenomena described by Rothert is found in the process of oösphere formation in *Vaucheria,* as described very carefully from living material by Oltmanns (39). Oltmanns confirms and amplifies the observations of Thuret, relative to zoöspore formation in *Vaucheria,* and Strasburger and Berthold, who studied the process of zoöspore and oögonium formation.

According to Oltmanns, just before the cell-division, which cuts off the oögonium from the parent filament, there is an extrusion of water from the protoplasmic mass within the oögonium; the extruded water forms a large vacuole at or below the base of the oögonium (figs. 8–10, pl. 6–7). This stage is comparable to the first contraction phase with its large central vacuole and the formation of radial furrows beginning the delimitation of the spores, as noted by Rothert in *Saprolegnia.* In the case of *Vaucheria,* cutting off of the oögonium is first initiated by what may be called the cleavage vacuole. The condition is similar to that found in columella formation in *Pilobolus crystallinus, Rhizopus nigricans, Phycomyces nitens, Sporodinia grandis, Mucor mucedo,* etc. Such basal vacuoles play the same rôle as the cleavage vacuoles which appear in the spore plasm of *Pilobolus, Phycomyces, Circinella,* etc.

The plasma membrane about the basal vacuole in *Vaucheria* is finally broken, the cell-sap escapes, and the oögonial protoplasm now expands; the basal plasma membrane of the oögonium and the plasma membrane of the filament are brought into close prox-

imity (fig. 9, pls. 6–7). This stage is to be compared also to the stage in the formation of the columella in sporangia of the Zygomycetes where the vacuoles flatten and fuse edge to edge. Later, in *Vaucheria,* a wall is formed between the two membranes and is seen to be convex toward the oögonium (fig. 11, pls. 6–7). The oösphere is now rounded up in the second contraction phase. The protoplasmic mass, which has heretofore conformed to the general outline of the oögonial wall, undergoes contraction until the rather globular or ovoid oösphere is formed. The ripe oösphere contains relatively few chloroplasts, but numerous oil globules, suggesting the chemical condensation processes which have accompanied the extrusion of cell-sap. Such illustrations show clearly that the process of spore formation, whether sexual or asexual, involves rather a marked series of contraction and expansion phases accompanied by metabolic changes in the protoplasm which result, in general, in the formation of reserve food products, but whose fundamental chemical nature is at present little known.

The process of spore formation may be much abbreviated as in *Sporodinia grandis,* whose spores are short lived and contain little reserve material, or it may be protracted as in *Pilobolus crystallinus* and *Synchytrium decipiens,* by the interpolation of an embryonic stage, in which the protospores increase in size, become multinucleated, ripen, form a wall, and enter a period of rest before they germinate by a tube in *Pilobolus* or by zoöspore formation in *Synchytrium.*

Swingle (50) attributes spore formation in sporangia as due to localized contractions of the protoplasm He does not believe that the nuclei directly influence contraction, but states: "The nuclei determine to some extent just what protoplasm shall constitute each individual spore."

Recently Harper (25) has suggested that the loss of water is probably least in the vicinity of the nuclei during the shrinking and condensation of the spore plasm, and that this might be a determining factor in the orientation of the cleavage furrows.

The failure to note the various contraction and expansion phases accompanying the formation of spores in the sporangia of algae and fungi has doubtless led to the erroneous conception of simul-

taneous cleavage as it still persists in the literature of spore formation.

While the method of the columella formation has been studied in relatively few Zygomycetes, the researches by Harper (24), Swingle (50), and myself have shown that the columella is not from the first a plane wall, which is subsequently pushed up into the sporangium, as is so often figured and described in textbooks on botany, but that it originates as a dome-shaped mass of vacuoles at the inner boundary of the spore plasm. Brefeld (7) observed that the columella was from the first dome-shaped. The vacuoles flatten in their radial axes, fuse edge to edge, and thus delimit the spore plasm from the columella plasm. In *Rhizopus nigricans, Pilobolus crystallinus,* and *Sporodinia grandis* a circular furrow cuts upward from the base of the sporangium to meet the cleft formed by the flattened vacuoles.

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EXPLANATION OF PLATES

All figures were drawn with the aid of the camera lucida, and with the Zeiss 1.8 mm. objective, N. A. 1.25; magnification about 1300 diameters.

PLATE 15

Sporodinia grandis

Fig. 1. Median vertical section of a sporangium showing cleavage complete on one side and first appearance of superficial cleavage furrows.

Fig. 2. Median vertical section of a sporangium showing columella-cleft completely formed and furrows passing upward through the spore plasm.

Fig. 3. Tangential section of a sporangium showing an advanced stage of cleavage and a large central mucorin crystal.

Fig. 4. Oblique section of a sporangium, spores have become polygonal by mutual pressure (expansion phase).

Fig. 5. Tangential section of a sporangium representing an early stage of cleavage.

Fig. 6. Median vertical section of a sporangium, columella-cleft and cleavage complete on one side of the sporangium and in an advanced stage on the other side.

PLATE 16

Fig. 7. *Sporodinia grandis.* Tangential section of a sporangium showing advanced stage of cleavage.

Circinella minor

Fig. 8. Tangential section of a sporangium showing branching furrows.

Fig. 9. Horizontal section of a sporangium; spore plasm cut into oblong to sausage-shaped protoplasmic masses which are somewhat concentrically arranged and are undergoing transverse segmentation.

Fig. 10. Section of a portion of a sporangium, protoplasmic masses are being cut up into 1–2 nucleated protospores.

Fig. 11. Vertical median section of a sporangium, somewhat later stage of cleavage than shown in figure 10.

Fig. 12. Oblique section of a sporangium about same stage as the last.

Fig. 13. Spores have become multinucleated and polygonal (expansion phase).

Fig. 14. Mature multinucleated spores.

Mucor racemosus

Fig. 15. Tangential section of a sporangium showing the beginning of two cleavage-furrows.

Fig. 16. Tangential section of a somewhat larger sporangium, the protoplasm is being cut up into irregular blocks.

Fig. 17. Tangential section of a sporangium, the irregular blocks of protoplasm are being cut up into spores, the nuclei are not well shown.

Fig. 18. Mature multinucleate spores.

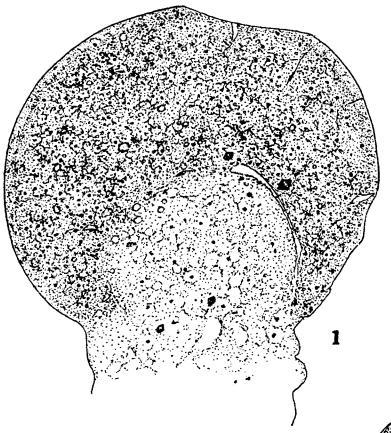
Mucor mucedo

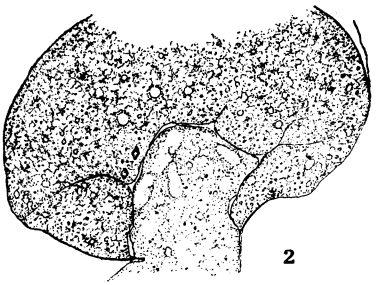
Fig. 19. Median vertical section of a sporangium, columella-cleft nearly complete on one side but in the early stage on the other side.

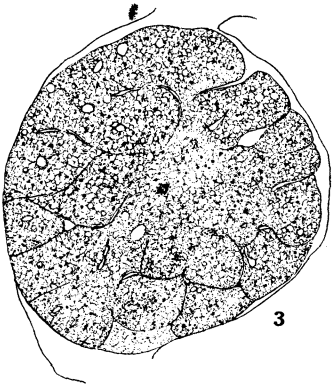
Fig. 20. Outline drawing of a section of a sporangium showing cleavage of spore-plasm into relatively large protoplasmic blocks.

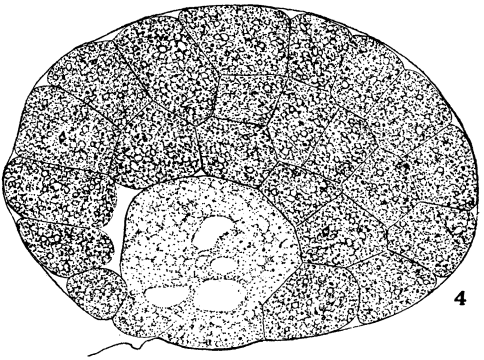
Fig. 21. Section of a sporangium showing spore-plasm cut up into uninucleate spore-initials.

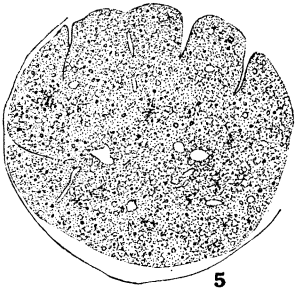
Fig. 22. Outline drawing of a median vertical section of a small sporangium showing polyhedral spore initials.

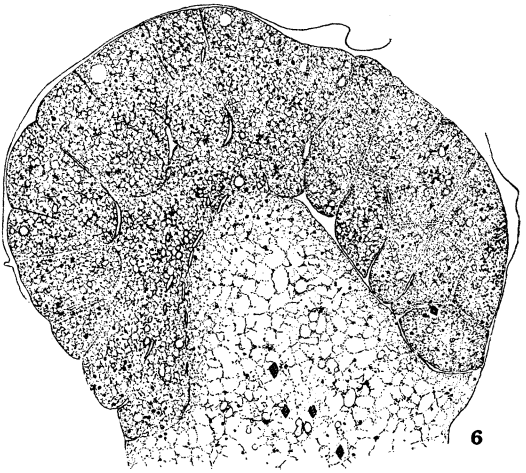












SPORODINIA GRANDIS

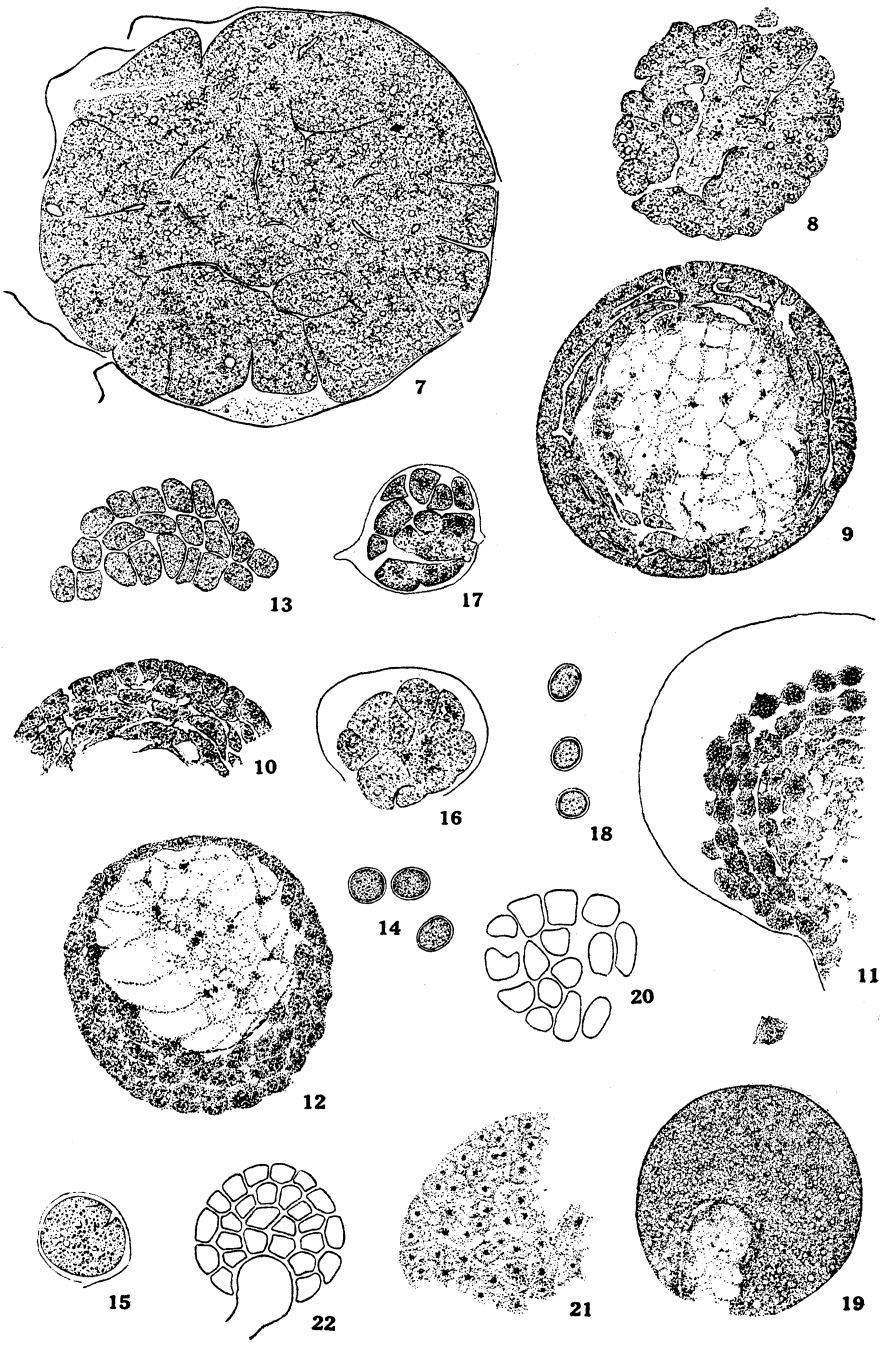


FIG. 7. SPORODINIA GRANDIS
FIG. 8–14. CIRCINELLA MINOR

FIG. 15–18. MUCOR RACEMOSUS
FIG. 19–22. MUCOR MUCEDO